

2.7. In Vivo Wounding Assay

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An abbreviated version of this protocol was published in Stem Cells International in May 2019
Epithelial-Mesenchymal Transition Promotes the Differentiation Potential of *Xenopus tropicalis* Immature Sertoli Cells
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Detailed protocol

2.7. In Vivo Wounding Assay.

*To analyze the wound homing capacity of XtiSCs, the wounding assay was performed as described [24] with modifications. Briefly, stage 51 or older (around 3-week-old) *X. tropicalis* larvae were anesthetized with 0.01% tricaine (MS-222) and put into a Petri dish with 6% Ficoll, 0.1x MMR, and 0.1% BSA.*

In our later experiments we found out that it is not necessary to put tadpoles to 6% Ficoll but 0,05 x MMR + gentamicin could be used instead for the microinjection environment.

Two hundred RFP-positive XtiSCs (40 nl) treated or untreated with CHIR99021 had been microinjected into larvae through dorsal blood vessels near the abdomen.

We used pneumatic microinjector IM-300 from Narishige for the cell microinjection and oil microinjector IM-55-2 for holding the anesthetized tadpole. The IM-300 allows microinjection so as aspiration of cell suspension. It is good to use fresh aspirated cell suspension after 5 injected tadpoles. Here is a figure showing the holding the tadpole and position of microinjection capillary before microinjection into dorsal vein



Just after microinjection, the distal third of the tail was wounded by #55 Forceps (Fine Science Tool). We just tweeke the the tail in the last third with forceps.

How to cite: (Readers should cite both the Bio-protocol preprint and the original research article where this protocol was used)

1. Krylov, V. (2021). 2.7. In Vivo Wounding Assay. Bio-protocol Preprint. bio-protocol.org/prep1187.
2. Nguyen, T. M. X., Vegrichtova, M., Tlapakova, T., Krulova, M. and Krylov, V. (2019). Epithelial-Mesenchymal Transition Promotes the Differentiation Potential of *Xenopus tropicalis* Immature Sertoli Cells. Stem Cells International 2019. DOI: [10.1155/2019/8387478](https://doi.org/10.1155/2019/8387478)

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